

CLAIMS

1. A plastid transformation vector for stably transforming a plastid genome, comprising, as operably-linked components, a first flanking sequence, at least one DNA sequence coding for a polypeptide suitable for remediating a contaminant compound, and a second flanking sequence, wherein a plant is stably transformed with said plastid transformation vector, and said plant is capable of phytoremediating a contaminant compound.

2. The vector of Claim 1, wherein said at least one DNA sequence coding for a polypeptide suitable for remediating a contaminant compound is a phytoremediation operon.

3. The vector of Claim 1 or 2 further comprising a regulatory sequence.

4. The vector of Claim 3, wherein said regulatory sequence comprises a promoter operative in said plastid genome.

5. The vector of Claim 4, wherein said promoter is 16srRNA.

6. The vector of Claim 3, wherein said regulatory sequence comprises a 3'untranslated region (UTR).

7. The vector of Claim 1, wherein the vector is competent for stably integrating in the plastid genome of a plant species and wherein the flanking DNA sequences are substantially homologous to sequences in a spacer region of said plastid genome, and wherein said flanking sequences are conserved in the plastid genome of said plant species.

8. The vector of Claim 7, wherein said spacer region is a transcriptionally active spacer region.

9. The vector of Claim 1, wherein the plastid genome is selected from the group consisting of chloroplast, chromoplast, amyloplast, proplastide, leucoplast and etioplast.

10. The vector of Claim 1 further comprising a DNA sequence encoding a selectable marker encoding an antibiotic-free selectable marker.

11. The vector of Claim 1, wherein said first flanking sequence is trnI, and wherein said second flanking sequence is trnA.

12. The vector of Claim 11, wherein trnI and trnA provide for homologous recombination to insert an operon coding for a protein suitable for inactivating a

contaminant compound into the spacer region in an inverted repeat region of a chloroplast genome.

13. The vector of Claim 1, wherein said operon is located in a single copy region of said plastid genome.

5 14. The vector of Claim 6, wherein said 3'UTR is a 3'UTR of psbA.

15. The vector of Claim 1, further comprising a DNA sequence encoding a selectable marker.

16. The vector of Claim 15, wherein said selectable marker is an antibiotic-free selectable marker.

10 17. The vector of Claim 16, wherein said antibiotic-free selectable marker is Betaine aldehyde dehydrogenase (BADH).

18. The vector of Claim 15, wherein said DNA sequence encoding a selectable marker encodes an antibiotic resistant selectable marker.

15 19. The vector of Claim 18, wherein said antibiotic resistant selectable marker is *aadA*.

20. A method for producing at least one DNA sequence coding for a protein suitable for inactivating a contaminant compound comprising:

integrating the plastid transformation vector of Claim 1 into the plastid genome of a plant cell;

20 growing said plant cell to thereby express said at least one heterologous DNA sequence coding for a protein suitable for inactivating a contaminant compound.

21. The method of Claim 20, wherein said at least one DNA sequence coding for a protein suitable for inactivating a contaminant compound is competent to phytoremediate a contaminant compound.

25 22. A plant stably transformed with the transformation vector of Claim 1.

23. A progeny of the plant of Claim 22.

24. A seed of the plant of Claim 22.

25 25. A plant part of the plant of Claim 22, comprising a plastid including said at least one heterologous DNA sequence coding for a protein suitable for inactivating a contaminant compound

30 26. The plant of Claim 22, wherein said plant further comprises at least one chloroplast transformed with the vector of Claim 1.

27. The plant of Claim 22, wherein said plant further comprises mature leaves transformed with the vector of Claim 1.

28. The plant of Claim 22, wherein said plant further comprises young leaves

5 transformed with the vector of Claim 1.

29. A plastid transformation vector for stably transforming a plastid genome, comprising, as operably-linked components, a first flanking sequence capable for integrating said plastid transformation vector into the plastid genome, an operon comprising *merA* and *merB* genes, and a second flanking sequence capable for
10 integrating said plastid transformation vector into the plastid genome.

30. The plastid transformation vector of Claim 29, wherein said first and second flanking sequences allow site-specific integration of the operon containing the *merA* and *merB* genes into an inverted repeat region of the plastid genome between *trnI* (tRNA Ile) and *trnA* (tRNA Ala) genes.

15 31. The plastid transformation vector of any one of claims 29 or 30, wherein said operon further comprises the *aadA* gene.

32. The plastid transformation vector of any one of claims 29-31, further comprising a 3' untranslated region (3'UTR) positioned downstream of the operon, and upstream of said second flanking sequence.

20 33. The plastid transformation vector of claim 32, wherein said 3'UTR is from a *psbA* chloroplast gene

34. A method of detoxifying mercury comprising the steps of:
integrating the vector of Claim 45 into a plastid genome of a plant cell
culturing said plant cell to express *merA* and *merB*,
25 exposing said plant cells to mercury.

35. The vector of Claim 2 wherein the operon is the *merAB* operon.

36. A plant cell comprising a plastid including an expression cassette, said expression cassette comprising as operably linked components, a promoter functional
30 in said plastid, an operon encoding a *merAB* operon, a transcription termination region, and DNA sequences flanking the expression cassette to facilitate stable integration of said expression cassette into a genome of said plastid by homologous recombination.

37. A plant cell comprising a plastid including an expression cassette, said expression cassette comprising as operably linked components, a promoter functional in said plastid, an operon encoding a phyto remediation operon, a transcription termination region, and DNA sequences flanking the expression cassette to facilitate
5 stable integration of said expression cassette into a genome of said plastid by homologous recombination.

38. The plastid transformation vector of claim 1, wherein said at least one DNA sequence coding for a polypeptide suitable for remediating a contaminant compound is a bacterial gene *onr*, encoding PETN reductase.

10 39. The plastid transformation vector of claim 1, wherein said at least one DNA sequence coding for a polypeptide suitable for remediating a contaminant compound is a cytochrome P450.

40. The plastid transformation vector of claim 1, wherein said at least one DNA molecule coding for a polypeptide suitable for remediating a contaminant
15 compound is a gene coding for phytochelatin synthase, wherein said phytochelatin synthase enables a transformed plant to sequester heavy metal ions.